PHILOSOPHICAL TRANSACTIONS A

royalsocietypublishing.org/journal/rsta

Research

Cite this article: Alexandrov DV, Nizovtseva IG. 2019 On the theory of crystal growth in metastable systems with biomedical applications: protein and insulin crystallization. *Phil. Trans. R. Soc. A* **377**: 20180214.
http://dx.doi.org/10.1098/rsta.2018.0214

Accepted: 1 November 2018

One contribution of 17 to a theme issue 'Heterogeneous materials: metastable and non-ergodic internal structures'.

Subject Areas:
complexity, mathematical modelling, applied mathematics, solid-state physics

Keywords:
crystal growth, nucleation, metastable liquid, phase transformation

Author for correspondence:
Irina G. Nizovtseva
e-mail: nizovtseva.irina@gmail.com

On the theory of crystal growth in metastable systems with biomedical applications: protein and insulin crystallization

Dmitri V. Alexandrov[1] and Irina G. Nizovtseva[2]

[1]Department of Theoretical and Mathematical Physics, Laboratory of Multi-Scale Mathematical Modeling, Ural Federal University, Ekaterinburg 620000, Russian Federation
[2]Physikalisch-Astronomische Fakultät, Friedrich-Schiller-Universität Jena, 07743 Jena, Germany

DVA, 0000-0002-6628-745X; IGN, 0000-0002-7766-7351

A generalized theory of nucleation and growth of crystals in a metastable (supercooled or supersaturated) liquid is developed taking into account two principal effects: the diffusion mechanism of the particle-size distribution function in the space of particle radii and the unsteady-state growth rates of individual crystals induced by fluctuations in external temperature or concentration field. A system of the Fokker–Planck and balance integro-differential equations is formulated and analytically solved in a parametric form for arbitrary nucleation kinetics and arbitrary growth rates of individual crystals. The particle-size distribution function and system metastability are found in an explicit form. The Weber–Volmer–Frenkel–Zel'dovich and Meirs kinetic mechanisms, as well as the unsteady-state growth rates of nuclei (Alexandrov & Alexandrova 2019 *Phil. Trans. R. Soc. A* **377**, 20180209 (doi:10.1098/rsta.2018.0209)), are considered as special cases. Some potential biomedical applications of the present theory for crystal growth from supersaturated solutions are discussed. The theory is compared with experimental data on protein and insulin crystallization (growth dynamics of the proteins lysozyme and canavalin as well as of bovine and porcine insulin is considered). The hat-shaped particle-size distribution functions for lysozyme and canavalin crystals as well as for bovine and porcine insulin are found.

THE ROYAL SOCIETY PUBLISHING

This article is part of the theme issue 'Heterogeneous materials: metastable and non-ergodic internal structures'.

1. Introduction

Traditionally, nucleation is usually associated with the initial stage of a phase transition occurring in a sufficiently pure metastable system, when the nuclei of the new phase arise as a result of fluctuations. The number of applied problems associated with this phenomenon is quite large: from crystal growth processes in industrial crystallizers to biological and medical applications (see, among others, [1–5]).

Ideas about the fluctuation formation of supercritical nuclei were first reported in the classical works of Volmer, Weber, Becker and Döring [6–8]. In these works, a kinetic problem was formulated for the case of stationary nucleation from a supersaturated vapour on the basis of ideas of the origin of nuclei owing to a large number of small fluctuations (in this case, the change in the dimensions of the nuclei was due to the addition or evaporation of individual atoms). Then Frenkel [9] showed that nucleation can occur even in a stable system as a result of equilibrium heterophase fluctuations. The next important step in the development of the theory of nucleation was the fundamental work of Zel'dovich [10]. In this paper, Zel'dovich showed that the crystal size distribution function has the form of the Fokker–Planck equation. In addition, the coefficients of this equation (two coefficients arise in terms proportional to the spatial derivatives) depend on the transition probabilities, which are determined by the change in the number of atoms (or molecules). On the basis of the distribution of equilibrium heterophase fluctuations, Zel'dovich obtained the relationship between the aforementioned coefficients (see also [11]). Therefore, the problem was reduced to finding only one coefficient—the rate of growth of the nuclei. This approach works particularly well for small nuclei. With the growth of large nuclei, there are a number of limitations connected with the fact that the distribution function of heterophase fluctuations is determined on the basis of the hypothesis of the homogeneity of the system surrounding the growing crystallite (in general, gradients of concentration and/or temperature always exist around the growing nucleus).

Summarizing the above, we note that the works of Volmer, Weber, Becker and Döring [6–8] form the basis of the classical theory of nucleation. Moreover, the equations determining the growth process are the Fokker–Planck-type equation for the particle-size distribution function and the mass balance and/or heat balance equation. An important fact is that the form of the Fokker–Planck kinetic equation can be different as a result of the physical nature of the nucleation process under consideration. For example, this equation is the first-order equation and has the form of a continuity equation in the case of neglecting 'diffusion' processes over the crystal size space. If this 'diffusion' is taken into account, this equation is the second-order equation with variable coefficients [11–13].

The process of phase transition in a metastable system is traditionally divided into several stages associated with the presence of characteristic relaxation times [12]. The initial stage of the phase transformation corresponds to large supersaturations (supercoolings), when, owing to nucleation, a large number of viable nuclei are formed (their initial size exceeds the critical one). This stage of the process is characterized by small sizes of nuclei located at large distances from each other. As this takes place, the level of metastability (supersaturation or supercooling) varies insignificantly. The description of the process kinetics at this stage is based on microscopic theory and depends to a large extent on the mechanisms of nucleation (for example, the nucleation rate). The next intermediate stage of the phase transition is characterized by two simultaneous processes: the growth of already existing particles, as well as the appearance (nucleation) of newly formed nuclei with dimensions exceeding the critical size. The level of metastability of the system at this stage decreases from the initial value to the value which is close to zero owing to the absorption of mass and/or the release of heat by the growing particles (crystals). The final

stage of the phase transition occurs when the nuclei reach macroscopic dimensions, the process of nucleation of new particles practically stops and supercooling (supersaturation) is low. At this stage, if the nuclei are smaller (larger) than the critical size, they dissolve (grow). In addition, the growth process of large particles is associated with the dissolution of smaller fractions of particles.

A complete system of equations describing the corresponding stage of the phase transition is a nonlinear integro-differential system. Note that there are no general methods for solving such systems of equations. Thus, for example, when describing the intermediate stage of the phase transformation, a quasi-stationary approximation is very often used for the growth of individual crystals [14–19]. We also note that an exact analytical description of the phase transition at the stage of Ostwald ripening is known only in the asymptotic case of large process times. In the classical works, Lifshitz & Slyozov [20,21] theoretically proved the existence of a stable asymptotic state of disperse systems, which is characterized by a universal distribution function. Then a series of theoretical papers followed, taking into account the various physical effects at the final stage of the phase transition (so, for instance, the initial distribution function at the final stage [22,23] and different mass transfer mechanisms [24,25] may be mentioned). However, until now the question of the system transition from the intermediate stage to its final state remains open. The solution of such problems due to nonlinearity of the integro-differential model of the process is carried out using cumbersome numerical schemes and methods [26,27].

Obtaining new results in various biochemical and biomedical research is hindered by the lack of a complete understanding of the mechanisms of nucleation and growth of crystals. For example, the slow dissolution rate of protein crystals is used to produce drugs such as insulin [28,29]. Note that the nucleation process controls the synthesis of various proteins (such as interferon-alpha and human growth hormone). Here, the following circumstance plays an important role. Suppose that the administered dose consists of a certain number of crystals. Then the release rate of the drug (duration of drug exposure) can be maintained for a longer time if the crystals are large enough (compared with the same dose of smaller crystals). To achieve such a crystal size distribution, the nucleation time needs to be small enough. This occurs when crystals evolve under conditions of practically unchanged metastability level (supersaturation). Other biochemical applications also take place. As an example, one can mention the pathological states associated with the formation of crystals or other solid aggregates in the human body. For example, the processes of crystallization of haemoglobin C and polymerization of haemoglobin S lead to a decrease in the plasticity of red blood cells and cause sickle cell anemia [30,31]. Another vital process responsible for the formation of cataracts is the phase transformation of proteins in the retina of the eye [32].

In this paper, we develop a generalized theory of nucleation and growth of nuclei at the intermediate stage of phase transformations in metastable solutions and melts, taking into account the arbitrary growth rates of crystals and the arbitrary kinetics of nucleation. The developed theory is applied to the description of crystallization of various biochemical substances.

2. The intermediate stage of phase transformations: a generalized set of governing equations

Let us consider a phase transition in a supercooled liquid or a supersaturated solution, where solid-phase particles arise as a result of nucleation and growth. The heat of phase transformation released by the growing crystals partially compensates the system supercooling ΔT, which varies from the initial value ΔT_0 to almost zero value with the passage of time t. In the case of supersaturated solutions, crystal growth occurs as a result of the absorption of the impurity concentration dissolved in the liquid. In this case, the supersaturation ΔC changes with time also from the initial value ΔC_0 to practically zero value. Let us especially emphasize that, when the supercooling (supersaturation) of the system goes to zero, the intermediate stage of phase transformation considered here passes to the final stage, which, in turn, should be

considered within the framework of the modified mathematical model. At this stage, such processes as Ostwald ripening, coagulation and disintegration of crystals can occur (see, among others, [33–38]). Taking this into account we describe here the intermediate stage only.

There are theoretical and experimental studies showing that the rate of particle growth in a metastable system undergoes random fluctuations [11,39,40]. In this case, the radius $r(t)$ of growing and fluctuating crystals is a random variable that satisfies the stochastic differential equation [41]

$$dr = V\,dt + \sqrt{2\tilde{D}}dW,$$

where V is the growth rate of a nucleus without fluctuations, $\tilde{D}$ is a function determining the rate of fluctuations (the coefficient of mutual Brownian diffusion) and W is the Wiener process. The distribution function $f(r,t)$ over the radii of growing crystals is described by the following equation of the Fokker–Planck type:

$$\frac{\partial f}{\partial t} + \frac{\partial}{\partial r}(Vf) = \frac{\partial}{\partial r}\left(\tilde{D}\frac{\partial f}{\partial r}\right), \quad r > r_*,\ t > 0, \tag{2.1}$$

where r_* represents the radius of critical crystals. Note that the right-hand side of equation (2.1) describes the 'diffusion' of the distribution function over the crystal size space.

Denoting the initial supercooling (supersaturation) of the liquid through ΔT_0 (ΔC_0), let us write the heat (mass) balance equation governing the dynamics of dimensionless metastability w as

$$w(t) = 1 - b\int_{r_*}^{\infty} r^3 f(r,t)\,\mathrm{d}r,\ t > 0. \tag{2.2}$$

Here, w and b in the cases of supercooled melts (sm) and supersaturated solutions (ss) can be expressed as

$$w(t) = \begin{cases} \dfrac{\Delta T(t)}{\Delta T_0}\ \text{(sm)}, \\ \dfrac{\Delta C(t)}{\Delta C_0}\ \text{(ss)}, \end{cases} \qquad b = \begin{cases} \dfrac{4\pi L_V}{3\rho_m C_m \Delta T_0}\ \text{(sm)}, \\ \dfrac{4\pi C_p}{3\Delta C_0}\ \text{(ss)}. \end{cases} \tag{2.3}$$

Here, $L_V = \rho_s L$, ρ_s is the density of the solid phase, L is the latent heat of phase transition, ρ_m and C_m represent the density and specific heat of the mixture, respectively, and C_p designates the concentration at saturation.

Let us choose the initial conditions (at $t = 0$) for the aforementioned equations in the form

$$f(r,0) = f_0(r), \quad w(0) = 1. \tag{2.4}$$

We assume that there are no crystals of infinitely large dimensions in the system, i.e. we have the boundary condition

$$f(\infty,t) = 0. \tag{2.5}$$

The flux of crystals that overcome the nucleation barrier is determined by the nucleation rate $I(w)$. Taking this into account, we get the boundary condition of the form

$$V(t)f(r_*,t) - \left(\tilde{D}\frac{\partial f}{\partial r}\right)_{r=r_*} = I(w). \tag{2.6}$$

Generally speaking, $I(w)$ should be determined on the basis of experimental data. Here, we develop the theory for arbitrary dependence $I(w)$. However, let us note two kinetics of nucleation frequently used in the literature

$$I = I_* \begin{cases} \exp\left[-p\left(\dfrac{\Delta T_0}{\Delta T}\right)^2\right]\ \text{(sm)}, \\ \exp\left[-p\ln^{-2}\left(\dfrac{C_l}{C_p}\right)\right]\ \text{(ss)}, \end{cases} \qquad I = I_* \begin{cases} (\Delta T)^p\ \text{(sm)}, \\ (\Delta C)^p\ \text{(ss)}, \end{cases} \tag{2.7}$$

where the pre-exponential factor I_* and parameter p are constants (see, for details, [17]). These nucleation kinetics are often called the Weber–Volmer–Frenkel–Zel'dovich (WVFZ) and Meirs

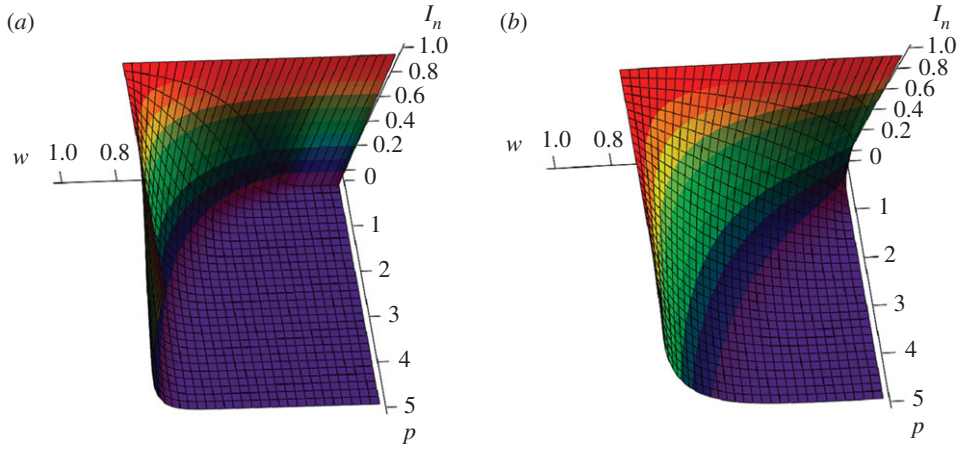

Figure 1. Relative nucleation rate I_n as a function of w and p for the WVFZ (*a*) and Meirs (*b*) nucleation kinetics in accordance with expressions (2.8).

kinetics. Let us especially note that parameters I_* and p in the WVFZ and Meirs nucleation mechanisms are different. Introducing $I_0 = I(\Delta T_0)$ and $I_0 = I(\Delta C_0)$ for 'sm' and 'ss' systems, one can rewrite the nucleation kinetics (2.7) as [12,13]

$$I(w) = I_0 \begin{cases} \exp\left[p\left(1 - w^{-2}\right)\right] \text{ (sm)}, \\ \exp\left[p\left(\ln^{-2}\left(1 + w_p^{-1}\right) - \ln^{-2}\left(1 + \dfrac{w}{w_p}\right)\right)\right] \text{ (ss)} \end{cases} \tag{2.8}$$

and

$$I(w) = I_0 \begin{cases} w^p \text{ (sm)}, \\ w^p \text{ (ss)}, \end{cases}$$

where $w_p = C_p/\Delta C_0$, $\Delta T = T_p - T_l$, $\Delta C = C_l - C_p$, T_p is the phase transition temperature, T_l is the current temperature and C_l is the current impurity concentration of the liquid system. Figure 1 compares two mechanisms of nucleation kinetics in the case of supercooled melts. It is easily seen that the relative nucleation rate $I_n = I/I_0$ shown as a function of two variables w and p represents a steeper function in the case of WVFZ nucleation kinetics.

It is important to note that an accurate determination of the coefficient $\tilde{D}$ is a complex problem of statistical physics. For simplicity, we employ below the following frequently used expression [42–44]:

$$\tilde{D} = d_1 V(t), \tag{2.9}$$

where d_1 is constant.

The integro-differential model (2.1)–(2.9) is solved below with allowance for arbitrary growth rates $V(t)$ of crystals and arbitrary nucleation kinetics $I(w)$.

3. The intermediate stage of phase transformations: analytical solutions

For convenience of solving the problem, we introduce dimensionless variables and parameters as follows:

$$s = \frac{r}{l_0},\ \tau = \frac{t}{t_0},\ F(s,\tau) = l_0^4 f(r,t),\ F_0(s) = l_0^4 f_0(r),\ U = \frac{Vt_0}{l_0},\ u_0 = \frac{d_1}{l_0}$$

and

$$l_0 = \left(\frac{\alpha_*}{I_0}\right)^{1/4},\ t_0 = \frac{l_0}{\alpha_*},\ x(\tau) = \int_0^\tau U(\bar{\tau})\, \mathrm{d}\bar{\tau},\ \alpha_* = \begin{cases} \beta_* \Delta T_0 \text{ (sm)}, \\ \beta_* \Delta C_0 \text{ (ss)}, \end{cases} \tag{3.1}$$

where β_* represents the kinetic coefficient entering in the growth rate of crystals (3.16). For the sake of simplicity, we neglect the critical radius r_*. The model equations (2.1)–(2.9) in dimensionless variables (3.1) become

$$\frac{\partial F}{\partial x}+\frac{\partial F}{\partial s}=u_0\frac{\partial^2 F}{\partial s^2},\quad s>0,\ x>0, \tag{3.2}$$

$$w(x)=1-b\int_0^\infty s^3F(s,x)\,\mathrm{d}s,\quad x>0, \tag{3.3}$$

$$F(s,0)=F_0(s),\quad w(0)=1, \tag{3.4}$$

$$F(\infty,x)=0 \tag{3.5}$$

and

$$F(0,x)-u_0\left(\frac{\partial F}{\partial s}\right)_{s=0}=J(w)=\frac{\exp\left(p\varphi(w)\right)}{U}, \tag{3.6}$$

where

$$\varphi(w)=\begin{cases}1-w^{-2}\ (\mathrm{sm}),\\ \ln^{-2}\left(1+w_p^{-1}\right)-\ln^{-2}\left(1+\dfrac{w}{w_p}\right)\ (\mathrm{ss}),\end{cases}\qquad \varphi(w)=\begin{cases}\ln w\ (\mathrm{sm}),\\ \ln w\ (\mathrm{ss})\end{cases}$$

in the cases of WVFZ and Meirs kinetics, respectively.

Applying the Laplace transform to equation (3.2) and keeping in mind the boundary conditions (3.4)–(3.6), we obtain

$$pF^*(s)-F_0(s)+\frac{\mathrm{d}F^*}{\mathrm{d}s}=u_0\frac{\mathrm{d}^2F^*}{\mathrm{d}s^2} \tag{3.7}$$

and

$$F^*(\infty)=0,\quad F^*(0)-u_0\left(\mathrm{d}F^*/\mathrm{d}s\right)_{s=0}=J^*. \tag{3.8}$$

Here $*$ denotes the Laplace transform with respect to x, and p is the Laplace variable. The solution of equation (3.7) with the boundary conditions (3.8) can be expressed as

$$\begin{aligned}F^*(s)=&\frac{J^*\exp\left(\alpha_1 s\right)}{1-u_0\alpha_1}-\frac{(1-u_0\alpha_2)\exp\left(\alpha_1 s\right)}{u_0(1-u_0\alpha_1)(\alpha_2-\alpha_1)}\int_0^\infty F_0\left(\bar{s}\right)\exp\left(-\alpha_2\bar{s}\right)\,\mathrm{d}\bar{s}\\&+\frac{1}{u_0(\alpha_2-\alpha_1)}\int_0^s F_0\left(\bar{s}\right)\exp\left(\alpha_1\left(s-\bar{s}\right)\right)\,\mathrm{d}\bar{s}\\&+\frac{1}{u_0(\alpha_2-\alpha_1)}\int_s^\infty F_0\left(\bar{s}\right)\exp\left(-\alpha_2\left(\bar{s}-s\right)\right)\,\mathrm{d}\bar{s},\end{aligned} \tag{3.9}$$

where

$$\alpha_1=\frac{1-\sqrt{1+4u_0p}}{2u_0}\quad\text{and}\quad\alpha_2=\frac{1+\sqrt{1+4u_0p}}{2u_0}.$$

Now taking into account the inverse Laplace transforms [45]

$$\begin{aligned}&\frac{\exp\left(-\sqrt{\alpha(p+a_1)}\right)}{a_2+\sqrt{p+a_1}}\to\exp\left(-a_1x\right)\left[\frac{1}{\sqrt{\pi x}}\exp\left(-\frac{\alpha}{4x}\right)-a_2\exp\left(a_2\sqrt{\alpha}+a_2^2x\right)\right.\\&\left.\times\operatorname{erfc}\left(\frac{\sqrt{\alpha}}{2\sqrt{x}}+a_2\sqrt{x}\right)\right],\\&\frac{1}{\sqrt{1+4u_0p}}\to\frac{1}{2\sqrt{\pi u_0x}}\exp\left(\frac{-x}{4u_0}\right),\quad F_1^*F_2^*\to\int_0^x F_1\left(\bar{x}\right)F_2\left(x-\bar{x}\right)\,\mathrm{d}\bar{x},\end{aligned}$$

where a_1, a_2 and α are positive constants, one can get the distribution function from expression (3.9) in the form

$$F(s,x)=\exp\left(\frac{s}{2u_0}\right)\int_0^x\frac{J(x-y)}{\sqrt{u_0}}\exp\left(-\frac{y}{4u_0}\right)\left[\frac{1}{\sqrt{\pi y}}\exp\left(-\frac{s^2}{4u_0y}\right)-\frac{1}{2\sqrt{u_0}}\exp\left(\frac{s}{2u_0}+\frac{y}{4u_0}\right)\operatorname{erfc}\left(\frac{s}{2\sqrt{u_0y}}+\frac{\sqrt{y}}{2\sqrt{u_0}}\right)\right]\mathrm{d}y+M(s,x), \tag{3.10}$$

where

$$\begin{aligned}M(s,x)=&\frac{1}{2\sqrt{\pi u_0x}}\int_0^s F_0(\bar{s})\exp\left[-\left(\frac{s-\bar{s}}{2\sqrt{u_0x}}-\frac{\sqrt{x}}{2\sqrt{u_0}}\right)^2\right]\mathrm{d}\bar{s}\\&+\frac{1}{2\sqrt{\pi u_0x}}\int_s^\infty F_0(\bar{s})\exp\left[-\left(\frac{\bar{s}-s}{2\sqrt{u_0x}}+\frac{\sqrt{x}}{2\sqrt{u_0}}\right)^2\right]\mathrm{d}\bar{s}\\&-\exp\left(\frac{s}{2u_0}\right)\int_0^\infty F_0(\bar{s})\exp\left(-\frac{\bar{s}}{2u_0}\right)\\&\times\left[\int_0^x\frac{Y(s,\bar{s},x-\bar{x})}{2\sqrt{\pi u_0\bar{x}}}\exp\left(-\frac{\bar{x}}{4u_0}\right)\mathrm{d}\bar{x}-Y(s,\bar{s},x)\right]\mathrm{d}\bar{s},\\Y(s,\bar{s},x)=&\frac{1}{2\sqrt{u_0}}\exp\left(-\frac{x}{4u_0}\right)\left[\frac{1}{\sqrt{\pi x}}\exp\left(-\frac{(s+\bar{s})^2}{4u_0x}\right)\right.\\&\left.-\frac{1}{2\sqrt{u_0}}\exp\left(\frac{s+\bar{s}}{2u_0}+\frac{x}{4u_0}\right)\operatorname{erfc}\left(\frac{s+\bar{s}}{2\sqrt{u_0x}}+\frac{\sqrt{x}}{2\sqrt{u_0}}\right)\right],\end{aligned}$$

and $J(x-y)=\exp[p\varphi(w(x-y))]/U(x-y)$. Note that our new solution (3.10) transforms to the previously known solution in the case $F_0=0$ [12,13]. If this is really the case, the function $M(s,x)$ vanishes. From the physical point of view this case describes the nucleation process with zero initial distribution function when crystals begin to nucleate and grow at $t>0$ (or $x>0$). The present distribution function (3.10) takes into account a non-zero initial distribution $f_0(r)$ (or $F_0(s)$).

Substitution of the distribution function (3.10) into the integral equation for the supercooling (supersaturation) (3.3) gives

$$w(x)=1-b\int_0^x J(x-y)h(y)\,\mathrm{d}y-\lambda(x), \tag{3.11}$$

where

$$\begin{aligned}\lambda(x)=&\,b\int_0^\infty s^3M(s,x)\,\mathrm{d}s,\\h(y)=&\frac{2}{\sqrt{\pi}}\left[\exp\left(-\frac{y}{4u_0}\right)\left(\frac{1}{2}u_0^{1/2}y^{5/2}+\frac{7}{2}u_0^{3/2}y^{3/2}-3u_0^{5/2}y^{1/2}\right)\right.\\&\left.+\sqrt{\pi}\left(\frac{9}{2}u_0y^2+3u_0^3+\frac{y^3}{2}\right)\right]-\operatorname{erfc}\left(\frac{\sqrt{y}}{2\sqrt{u_0}}\right)\left[6u_0^3+\frac{9}{2}u_0y^2+\frac{y^3}{2}\right].\end{aligned}$$

If we consider the inverse function $y(w)$, (3.11) becomes

$$w=1-b\int_1^w J(w_1)h[y(w_1)]\frac{\mathrm{d}y}{\mathrm{d}w_1}\,\mathrm{d}w_1-\lambda(x(w)). \tag{3.12}$$

Now differentiating (3.12) with respect to variable w, we come to

$$\frac{\mathrm{d}w}{\mathrm{d}x}=-\frac{b\exp\left[p\varphi(w)\right]h(x)}{U(w,\tau)}-\frac{\mathrm{d}\lambda}{\mathrm{d}x}. \tag{3.13}$$

Next differentiating $x(\tau)$ from (3.1), we obtain

$$\frac{\mathrm{d}\tau}{\mathrm{d}x} = \frac{1}{U(w,\tau)}. \tag{3.14}$$

Equations (3.13) and (3.14) must be solved under the following initial conditions:

$$w = 1, \quad \tau = 0 \quad \text{and} \quad x = 0. \tag{3.15}$$

It is important to note that expressions (3.13)–(3.15) represent the Cauchy problem, which determines the parametric dependencies $w(x)$ and $\tau(x)$ for arbitrary growth rates $U(w,\tau)$, arbitrary nucleation kinetics, and arbitrary initial distribution function $F_0(s)$. As this takes place, the particle-size distribution function is given by expression (3.10). Let us especially note that the analytical solutions (3.10), (3.13)–(3.15) generalize previously known solutions obtained for some special cases in [12,13].

The growth rate of crystals V is a function of time and different heat and mass transfer parameters if crystals grow in supercooled and supersaturated liquids. The analytical theories developed for the thermally controlled growth [46], diffusionally controlled growth [47] and crystal growth in binary melts [48] give the following expressions for dimensional (V) and dimensionless (U) velocities

$$\left.\begin{aligned} V &= \begin{cases} \beta_* \Delta T\,(1 - \sigma_T \Delta T t) \ \text{(sm)}, \\ \beta_* \Delta C\,(1 + \sigma_C \Delta C t) \ \text{(ss)}, \end{cases} \qquad U = \begin{cases} w\,(1 - \chi_T w\tau) \ \text{(sm)}, \\ w\,(1 + \chi_C w\tau) \ \text{(ss)}, \end{cases} \\ \text{and} \qquad \sigma_T &= \frac{\beta_*^2 \rho_s L}{\lambda_l}, \quad \sigma_C = \frac{\beta_*^2 (1-k) C_p}{D}, \quad \chi_T = \sigma_T \Delta T_0 t_0, \quad \chi_C = \sigma_C \Delta C_0 t_0, \end{aligned}\right\} \tag{3.16}$$

where λ_l is the thermal conductivity, k is the partition coefficient and D is the diffusion coefficient. It is important to note that U is a composed function of τ, i.e. $U(\tau) = U(w(\tau),\tau)$. An important point is that expressions (3.16) derived for crystal growth in metastable one-component liquids take into account the non-stationary fluctuations of the temperature (concentration) field caused by the release of latent heat of crystallization (absorption and displacement of the dissolved impurity) on the surfaces of growing spherical particles. Specifically, we note that, if it is necessary to describe the growth rates of crystals more accurately, corrections of higher orders of smallness can be taken into account (for details, see [46–48]). The first term $U = w$ on the right-hand side of expression (3.16) represents the growth processes in steady-state temperature (concentration) fields and does not describe some fluctuations in the particle growth rates.

In concluding this section, we note an analytic solution of equations (3.13) and (3.14) in an explicit form in the simplest case $U = w$. Keeping in mind this approximation, we have

$$\int_1^w \bar{w} \exp[-p\varphi(\bar{w})]\,\mathrm{d}\bar{w} = H(x) \equiv -b\int_0^x h(y)\,\mathrm{d}y, \quad \tau(x) = \int_0^x \frac{\mathrm{d}\bar{x}}{w(\bar{x})}, \tag{3.17}$$

where $\varphi(w)$ depends on the mechanism of nucleation kinetics. In the case of the Meirs mechanism, the integral on the left-hand side of expression (3.17) can be evaluated in an explicit form,

$$w(x) = \begin{cases} \left[(2-p)H(x) + 1\right]^{1/(2-p)}, & p \neq 2, \\ \exp\left[H(x)\right], & p = 2. \end{cases} \tag{3.18}$$

In this simple case, the analytical solutions are given by expressions (3.10) and (3.18) in a parametric form (with parameter x).

The Cauchy problem (3.13)–(3.15) can be rewritten to determine an explicit dependence of the system metastability w of dimensionless time τ if U is a function of w only, i.e. if $U = w$ (χ_T or χ_C

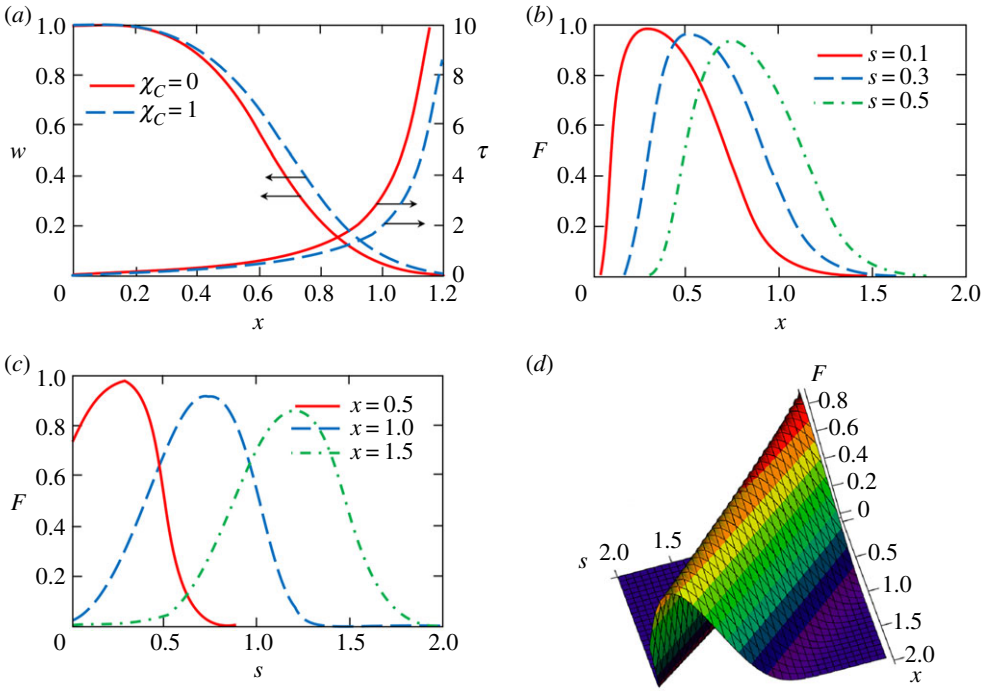

Figure 2. (*a*) The dimensionless supercooling (supersaturation) w (scale of values on the left) and the dimensionless time τ (scale of values on the right) as functions of the modified time x. (*b*,*c*) The dimensionless distribution function F versus the modified time x and dimensionless radius s, respectively. (*d*) The distribution function F versus s and x. The physical parameters used for calculations are [12]: $b = 14.92$, $u_0 = 10^{-2}$ and $p = 2.2$. Here, the Meirs kinetic mechanism is taken into account and $\chi_C = 0$ in (*b*–*d*).

are small enough) according to the main contribution on the right-hand side of expression (3.16). If this is really the case, we come to the following Cauchy problem:

$$\left.\begin{aligned} &\frac{\mathrm{d}x}{\mathrm{d}w} = \Sigma(w,x), \quad x=0,\ w=1 \\ \text{and}\quad &\Sigma(w,x) = -\frac{w}{b\exp\left[p\varphi(w)\right]h(x)+\lambda'(x)w},\ \lambda'(x) = \frac{\mathrm{d}\lambda}{\mathrm{d}x}. \end{aligned}\right\} \tag{3.19}$$

The initial value problem (3.19) enables us to find the explicit function $x(w)$ and its derivative $x'(w)$. Then an explicit dependence between w and τ can be easily found from (3.14) and (3.15) in the form of the inverse function

$$\tau(w) = \int_1^w \frac{x'(\bar{w})}{\bar{w}}\,\mathrm{d}\bar{w}. \tag{3.20}$$

This method allows us to eliminate parameter x from the solution of the problem.

Figure 2 illustrates the parametric solution (3.10), (3.13)–(3.15) for a metastable system that did not initially contain any solid crystals ($F_0 = 0$). The supercooling (supersaturation) w of a metastable liquid decreases and the dimensionless time τ increases with increasing the modified time x (figure 2*a*). In other words, w is a decreasing function of the real crystallization time τ as a result of the release of the latent heat of crystallization (absorption of the dissolved impurity) by the growing crystals. A time evolution of the distribution function F for crystals of a given size s is shown in figure 2*b*. Here, one can see that the bell-shaped distribution becomes lower with increasing size s of crystals. The particle-size distribution function F at fixed values of parameter x (modified time) is illustrated in figure 2*c*. A bell-shaped distribution moves to the right and

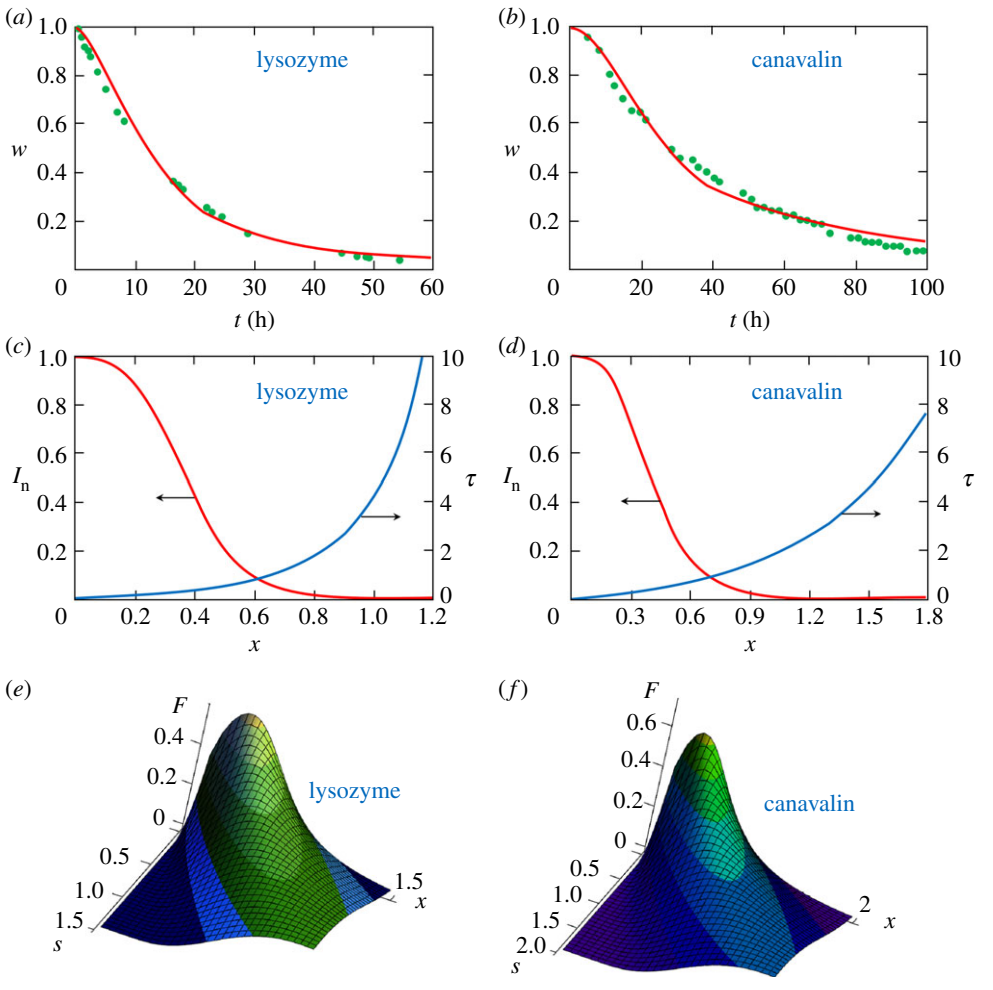

Figure 3. Dimensionless supersaturation w versus time t (*a*,*b*), relative nucleation rate I_n and time τ versus modified time x (*c*,*d*) and crystal-size distribution function versus dimensionless radius s and modified time x (*e*,*f*) for lysozyme and canavalin crystallization. The theory under consideration is compared with experimental data (filled circles) [50,51] in (*a*,*b*). Parameters used in calculations are: $b = 5, p = 2.4, u_0 = 0.3$ (lysozyme) and $b = 3, p = 3.4, u_0 = 0.3$ (canavalin).

becomes lower as time x (and also τ) increases. Figure 2*d* illustrates a three-dimensional plot of the distribution function and generalizes its behaviour shown in figure 2*b*,*c*.

4. Biomedical applications

The theory developed in this paper plays a decisive role in describing the nucleation of a new phase and growth of crystals during aggregation in colloids and magnetic fluids, in the production of insulin, proteins and other vital chemical substances. So, for instance, an important part of investigations of protein growth is carried out using protein lysozyme, most often extracted from hen egg white. The importance of studying this enzyme is connected with the fact that it hydrolyses polysaccharides in bacterial cell walls. It can be used as an antiseptic and as a food addition too [49]. Note that the rate of decay of the protein supersaturation in crystallizing solutions of chicken egg-white lysozyme was studied by Kim *et al.* [50] when the crystalline protein is more stable than the dissolved protein. The growth dynamics of another important protein, canavalin (the jack bean storage protein), was studied by Caraballo *et al.*

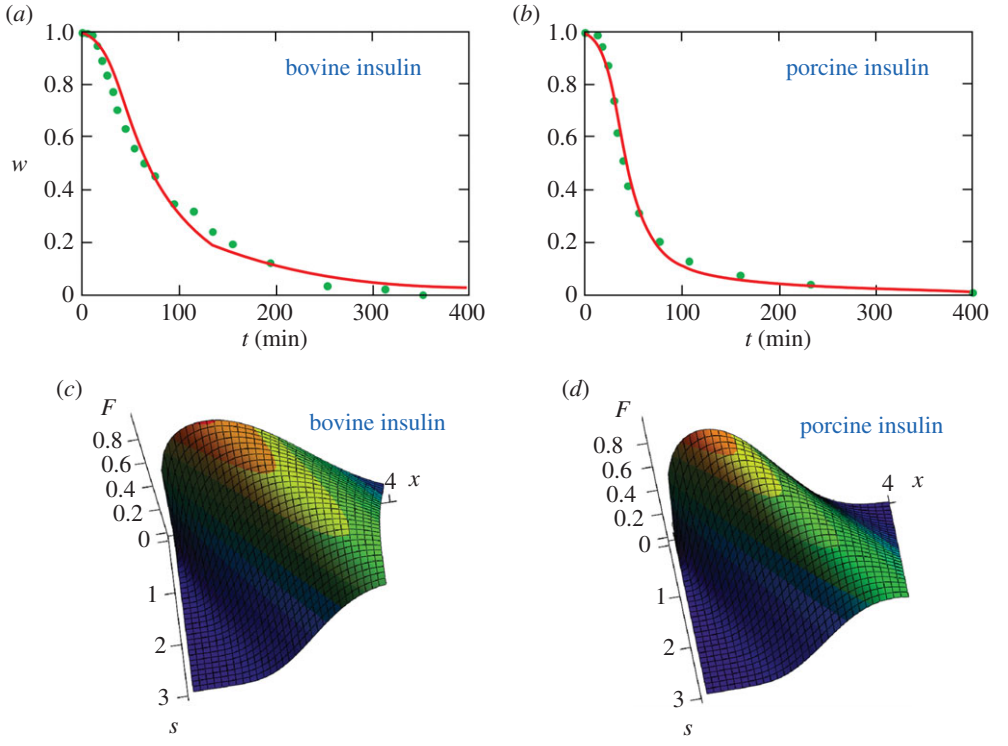

Figure 4. Dimensionless supersaturation w versus time t (*a*,*b*), and crystal-size distribution function versus dimensionless radius s and modified time x (*c*,*d*) for bovine and porcine insulin. The theory under consideration is compared with experimental data (filled circles) [52,53] in (*a*,*b*). Parameters used in calculations are: $b = 0.02$, $p = 2.3$, $u_0 = 0.3$ (bovine) and $b = 0.08$, $p = 1.9$, $u_0 = 0.3$ (porcine).

[51]. We compare below the theory under consideration and experimental data on protein crystallization [50,51].

Figure 3*a*,*b* shows a decreasing behaviour of the relative supersaturation with time. Note that the Meirs mechanism was chosen to describe the nucleation rate as a function of supersaturation. Our analytical solution enables us to find the relative nucleation rate of lysozyme and canavalin crystals as a function of modified time x (figure 3*c*,*d*). The dependencies between the real (τ) and modified (x) time variables are shown here too (scale of values on the right). Comparing these functions, we can conclude that the relative nucleation rate I_n decreases with increasing process time τ (or with decreasing system supersaturation w as a result of the evolution of protein crystals). A hat-shaped particle-size distribution function for lysozyme and canavalin crystals is shown in figure 3*e*,*f*. If we fix, for example, a moment in time (x or τ), the distribution function becomes bell-shaped owing to the influence of the so-called diffusion term entering on the right-hand side of the Fokker–Planck equation (2.1). If we fix a particular size of crystals (variable s), the distribution function also represents a bell-shaped curve that evolves with time in a manner similar to that shown in figure 2*b*.

Let us now compare the theory under consideration with important experimental data on insulin crystal growth reported by Schlichtkrull [52,53]. Figure 4*a*,*b* demonstrates the dimensionless supersaturation as a function of time for bovine and porcine insulin. As is easily seen, the theory is in good agreement with experiments. Small deviations between theoretical curves and experimental dependencies can be explained, for example, by the fact that the shape of both the bovine and porcine insulin crystals differs from spheres. So, insulin crystals can take the shape of cubes, dodecahedrons and rhombohedrons [54]. Therefore, with a more rigorous

study of the growth dynamics of insulin crystals, their real non-spherical growth shape must be taken into account. Non-sphericity of the form will, in turn, change the law of growth rate of insulin crystals. In other words, the growth rate of such crystals can be a composed function of supersaturation and time. One of the approximations is the use of the power-law dependence of the crystal growth rate on supersaturation [55]. A hat-shaped particle-size distribution function for bovine and porcine insulin crystals is illustrated in figure 4*c*,*d*. Here, one can see that both functions look similar. However, the distribution function shown in figure 4*d* demonstrates a more abrupt behaviour than the function plotted in figure 4*c*.

Concluding this section let us emphasize that the present theory of nucleation and growth of crystals in metastable liquids at the intermediate stage is suitable for the description of phase transition dynamics in the case of protein and insulin crystallization.

5. Conclusion

In the present paper, a generalized theory of crystal growth in a metastable one-component liquid at the intermediate stage of a phase transformation process is developed. A parametric solution for arbitrary nucleation kinetics and arbitrary growth rate of spherical crystals is constructed. The obtained analytical solution transforms to the previously known solutions in more simple cases of crystal growth. An explicit solution is also found when the growth rate depends on the system supersaturation only. The obtained analytical solutions are in good agreement with experiments on non-stationary nucleation and growth of insulin and protein crystals. The obtained particle-size distribution function (3.10) represents an initial distribution of crystals in a metastable liquid for the theoretical description of the concluding stage of a phase transformation process [35,56]. For a more accurate description of the process of crystal growth, it is interesting to generalize the present theoretical approach to describe the growth of non-spherical crystals, just as was done in describing the growth of ellipsoidal particles in metastable magnetic liquids [57]. It is also of interest to make a theoretical generalization of the bulk and directional crystallization for modelling solidification processes in the presence of a mushy layer (two-phase region) that is widely encountered in various fields of physics [58–64].

Data accessibility. This article has no additional data.

Authors' contributions. All authors contributed equally to the present research article.

Competing interests. We declare we have no competing interests.

Funding. This work was supported by the Russian Science Foundation (grant no. 18-19-00008).

References

1. Herlach D, Galenko P, Holland-Moritz D. 2007 *Metastable solids from undercooled melts*. Amsterdam, The Netherlands: Elsevier.
2. Kelton KF, Greer AL. 2010 *Nucleation in condensed matter: applications in materials and biology*. Amsterdam, The Netherlands: Elsevier.
3. Dubrovskii VG. 2014 *Nucleation theory and growth of nanostructures*. Berlin, Germany: Springer.
4. Alexandrov DV. 2014 Nucleation and crystal growth kinetics during solidification: the role of crystallite withdrawal rate and external heat and mass sources. *Chem. Eng. Sci.* **117**, 156–160. (doi:10.1016/j.ces.2014.06.012)
5. Makoveeva EV, Alexandrov DV. 2018 A complete analytical solution of the Fokker-Planck and balance equations for nucleation and growth of crystals. *Phil. Trans. R. Soc. A* **376**, 20170327. (doi:10.1098/rsta.2017.0327)
6. Volmer M, Weber A. 1926 Keimbildung in übersättigten gebilden. *Z. Phys. Chem.* **119**, 277–301. (doi:10.1515/zpch-1926-11927)
7. Volmer M. 1939 *Kinetik der Phasenbildung*. Dresden, Germany: Steinkopf.
8. Becker R, Döring W. 1935 Kinetische behandlung der keimbildung in übersättigten dämpfern. *Ann. Phys.* **24**, 719–752.
9. Frenkel J. 1955 *Kinetic theory of liquids*. New York, NY: Dover.

10. Zel'dovich JB. 1942 On the theory of formation of new phases: cavitation. *J. Exp. Theor. Phys.* **12**, 525–538.
11. Lifshitz EM, Pitaevskii LP. 1981 *Physical kinetics*. Oxford, UK: Pergamon.
12. Alexandrov DV, Nizovtseva IG. 2014 Nucleation and particle growth with fluctuating rates at the intermediate stage of phase transitions in metastable systems. *Proc. R. Soc. A* **470**, 20130647. (doi:10.1098/rspa.2013.0647)
13. Alexandrov DV. 2014 On the theory of transient nucleation at the intermediate stage of phase transitions. *Phys. Lett. A* **378**, 1501–1504. (doi:10.1016/j.physleta.2014.03.051)
14. Buyevich YuA, Mansurov VV. 1990 Kinetics of the intermediate stage of phase transition in batch crystallization. *J. Cryst. Growth* **104**, 861–867. (doi:10.1016/0022-0248(90)90112-X)
15. Buyevich YuA, Natalukha IA. 1994 Unsteady processes of combined polymerization and crystallization in continuous apparatuses. *Chem. Eng. Sci.* **49**, 3241–3247. (doi:10.1016/0009-2509(94)E0052-R)
16. Barlow DA. 2009 Theory of the intermediate stage of crystal growth with applications to protein crystallization. *J. Cryst. Growth* **311**, 2480–2483. (doi:10.1016/j.jcrysgro.2009.02.035)
17. Alexandrov DV, Malygin AP. 2013 Transient nucleation kinetics of crystal growth at the intermediate stage of bulk phase transitions. *J. Phys. A: Math. Theor.* **46**, 455101. (doi:10.1088/1751-8113/46/45/455101)
18. Alexandrov DV. 2014 Nucleation and crystal growth in binary systems. *J. Phys. A: Math. Theor.* **47**, 125102. (doi:10.1088/1751-8113/47/12/125102)
19. Alexandrov DV. 2014 Nucleation and growth of crystals at the intermediate stage of phase transformations in binary melts. *Phil. Mag. Lett.* **94**, 786–793. (doi:10.1080/09500839.2014.977975)
20. Lifshitz IM, Slyozov VV. 1959 Kinetics of diffusive decomposition of supersaturated solid solutions. *Sov. Phys.–JETP* **8**, 331–339.
21. Lifshitz IM, Slyozov VV. 1961 The kinetics of precipitation from supersaturated solid solutions. *J. Phys. Chem. Solids* **19**, 35–50. (doi:10.1016/0022-3697(61)90054-3)
22. Slezov VV. 1978 Formation of the universal distribution function in the dimension space for new-phase particles in the diffusive decomposition of the supersaturated solid solution. *J. Phys. Chem. Solids* **39**, 367–374. (doi:10.1016/0022-3697(78)90078-1)
23. Alexandrov DV. 2015 On the theory of Ostwald ripening: formation of the universal distribution. *J. Phys. A: Math. Theor.* **48**, 035103–035100. (doi:10.1088/1751-8113/48/3/035103)
24. Slezov VV, Sagalovich VV, Tanatarov LV. 1978 Theory of diffusive decomposition of supersaturated solid solution under the condition of simultaneous operation of several mass-transfer mechanisms. *J. Phys. Chem. Solids* **39**, 705–709. (doi:10.1016/0022-3697(78)90002-1)
25. Alexandrov DV. 2016 On the theory of Ostwald ripening in the presence of different mass transfer mechanisms. *J. Phys. Chem. Solids* **91**, 48–54. (doi:10.1016/j.jpcs.2015.12.005)
26. Madras G, McCoy BJ. 2002 Transition from nucleation and growth to Ostwald ripening. *Chem. Eng. Sci.* **57**, 3809–3818. (doi:10.1016/S0009-2509(02)00313-5)
27. Uwaha M, Koyama K. 2010 Transition from nucleation to ripening in the classical nucleation model. *J. Cryst. Growth* **312**, 1046–1054. (doi:10.1016/j.jcrysgro.2010.01.017)
28. Brange J. 1987 *Galenics of insulin*. Berlin, Germany: Springer.
29. Long MM, Bishop JB, Nagabhushan TL, Reichert P, Smith GD, DeLucas LJ. 1996 Protein crystal growth in microgravity review of large scale temperature induction method: bovine insulin, human insulin and human alpha interferon. *J. Cryst. Growth* **168**, 233–243. (doi:10.1016/0022-0248(96)00325-9)
30. Hirsch RE, Raventos-Suarez C, Olson JA, Nagel RL. 1985 Ligand state of intraerythrocytic circulating HbC crystals in homozygote CC patients. *Blood* **66**, 775–777.
31. Ferrone FA, Hofrichter J, Eaton WA. 1985 Kinetics of sickle hemoglobin polymerization. II. A double nucleation mechanism. *J. Mol. Biol.* **183**, 611–631. (doi:10.1016/0022-2836(85)90175-5)
32. Berland CR, Thurston GM, Kondo M, Broide ML, Pande J, Ogun O, Benedek GB. 1992 Solid-liquid phase boundaries of lens protein solutions. *Proc. Natl Acad. Sci. USA* **89**, 1214–1218. (doi:10.1073/pnas.89.4.1214)
33. Hunt JR. 1982 Self-similar particle-size distributions during coagulation: theory and experimental verification. *J. Fluid Mech.* **122**, 169–185. (doi:10.1017/S0022112082002158)

34. Simons S. 1996 On steady-state solutions of the coagulation equation. *J. Phys. A: Math. Gen.* **29**, 1139–1140. (doi:10.1088/0305-4470/29/5/026)
35. Slezov VV. 2009 *Kinetics of first-order phase transitions*. Weinheim, Germany: Wiley-VCH.
36. Alyab'eva AV, Buyevich YuA, Mansurov VV. 1994 Evolution of a particulate assemblage due to coalescence combined with coagulation. *J. Phys. II France* **4**, 951–957. (doi:10.1051/jp2:1994176)
37. Alexandrov DV. 2016 Kinetics of particle coarsening with allowance for Ostwald ripening and coagulation. *J. Phys. Condens. Matter* **28**, 035102. (doi:10.1088/0953-8984/28/3/035102)
38. Alexandrov DV. 2018 The steady-state solutions of coagulation equations. *Int. J. Heat Mass Trans.* **121**, 884–886. (doi:10.1016/j.ijheatmasstransfer.2018.01.069)
39. Zumstein RC, Rousseau RW. 1987 Growth rate dispersion by initial growth rate distributions and growth rate fluctuations. *AIChE J.* **33**, 121–129. (doi:10.1002/aic.690330114)
40. Randolph A, Larson M. 1988 *Theory of particulate processes*. San Diego, CA: Academic Press.
41. Gardiner CW. 1983 *Handbook on stochastic methods: for physics, chemistry and natural sciences*. Berlin, Germany: Springer.
42. Melikhov IV, Belousova TYa, Rudnev NA, Bludov NT. 1974 Fluctuation in the rate of growth of monocrystals. *Kristallografiya* **19**, 1263–1268.
43. Buyevich YuA, Ivanov AO. 1993 Kinetics of phase separation in colloids. II. Non-linear evolution of a metastable colloid. *Physica A* **193**, 221–240. (doi:10.1016/0378-4371(93)90027-2)
44. Alexandrov DV, Malygin AP. 2014 Nucleation kinetics and crystal growth with fluctuating rates at the intermediate stage of phase transitions. *Model. Simul. Mater. Sci. Eng.* **22**, 015003. (doi:10.1088/0965-0393/22/1/015003)
45. Ditkin VA, Prudnikov AP. 1965 *Integral transforms and operational calculus*. New York, NY: Pergamon Press.
46. Alexandrov DV. 2018 Nucleation and evolution of spherical crystals with allowance for their unsteady-state growth rates. *J. Phys. A: Math. Theor.* **51**, 075102. (doi:10.1088/1751-8121/aaa5b7)
47. Alexandrov DV, Nizovtseva IG, Alexandrova IV. 2019 On the theory of nucleation and nonstationary evolution of a polydisperse ensemble of crystals. *Int. J. Heat Mass Trans.* **128**, 46–53. (doi:10.1016/j.ijheatmasstransfer.2018.08.119)
48. Alexandrov DV, Alexandrova IV. 2019 On the theory of the unsteady-state growth of spherical crystals in metastable liquids. *Phil. Trans. R. Soc. A* **377**, 20180209. (doi:10.1098/rsta.2018.0209)
49. Rosenberger F, Vekilov PG, Muschol M, Thomas BR. 1996 Nucleation and crystallization of globular proteins—what we know and what is missing. *J. Cryst. Growth* **168**, 1–27. (doi:10.1016/0022-0248(96)00358-2)
50. Kim YW, Barlow DA, Caraballo KG, Baird JK. 2003 Kinetics of supersaturation decay in the crystallization of lysozyme. *Mol. Phys.* **101**, 2677–2686. (doi:10.1080/00268970310001358 52)
51. Caraballo KG, Baird JK, Ng JD. 2006 Comparison of the crystallization kinetics of canavalin and lysozyme. *Cryst. Growth Des.* **6**, 874–880. (doi:10.1021/cg058016u)
52. Schlichtkrull J. 1957 Insulin crystals. V. The nucleation and growth of insulin crystals. *Acta Chem. Scand.* **11**, 439–460. (doi:10.3891/acta.chem.scand.11-0439)
53. Schlichtkrull J. 1957 Insulin crystals. VII. The growth of insulin crystals. *Acta Chem. Scand.* **11**, 1248–1256. (doi:10.3891/acta.chem.scand.11-1248)
54. Ootaki M, Endo S, Sugawara Y, Takahashi T. 2009 Crystal habits of cubic insulin from porcine pancreas and evaluation of intermolecular interactions by macrobond and EET analyses. *J. Cryst. Growth* **311**, 4226–4234. (doi:10.1016/j.jcrysgro.2009.07.025)
55. Barlow DA. 2017 Theory of the intermediate stage of crystal growth with applications to insulin crystallization. *J. Cryst. Growth* **470**, 8–14. (doi:10.1016/j.jcrysgro.2017.03.053)
56. Alexandrov DV. 2015 Relaxation dynamics of the phase transformation process at its ripening stage. *J. Phys. A: Math. Theor.* **48**, 245101. (doi:10.1088/1751-8113/48/24/245101)
57. Ivanov AO, Zubarev AYu. 1998 Non-linear evolution of a system of elongated drop-like aggregates in a metastable magnetic fluid. *Physica A* **251**, 348–367. (doi:10.1016/S0378-4371(97)00561-X)
58. Martin S, Kauffman P. 1974 The evolution of under-ice melt ponds, or double diffusion at the freezing point. *J. Fluid Mech.* **64**, 507–527. (doi:10.1017/S0022112074002527)

59. Mansurov VV. 1990 The nonlinear dynamics of solidification of a binary melt with a nonequilibrium mushy region. *Math. Comput. Model.* **14**, 819–821. (doi:10.1016/0895-7177(90)90296-Y)
60. Kerr RC, Woods AW, Worster MG, Huppert HE. 1990 Solidification of an alloy cooled from above. Part I. Equilibrium growth. *J. Fluid Mech.* **216**, 323–342. (doi:10.1017/S0022112090000453)
61. Aseev DL, Alexandrov DV. 2006 Unidirectional solidification with a mushy layer. The influence of weak convection. *Acta Mater.* **54**, 2401–2406. (doi:10.1016/j.actamat.2006.01.039)
62. Alexandrov DV, Ivanov AA. 2009 Solidification of a ternary melt from a cooled boundary, or nonlinear dynamics of mushy layers. *Int. J. Heat Mass Trans.* **52**, 4807–4811. (doi:10.1016/j.ijheatmasstransfer.2009.05.029)
63. Alexandrov DV, Malygin AP. 2011 Convective instability of directional crystallization in a forced flow: the role of brine channels in a mushy layer on nonlinear dynamics of binary systems. *Int. J. Heat Mass Trans.* **54**, 1144–1149. (doi:10.1016/j.ijheatmasstransfer.2010.11.008)
64. Huguet L, Alboussière T, Bergman MI, Deguen R, Labrosse S, Lesoeur G. 2016 Structure of a mushy layer under hypergravity with implications for Earth's inner core. *Geophys. J. Int.* **204**, 1729–1755. (doi: 10.1093/gji/ggv554)